DNMT Activity / Inhibition Assay

convenient assay to screen for DNA methyltransferase activity

Active Motif's DNMT Activity / Inhibition Assay is a fast, user-friendly assay to simplify the measurement of DNA methyltransferase activity or the efficacy of DNMT inhibitors without the need for radioisotopes or expensive equipment. It can be used to study recombinant DNMT enzymes or nuclear extract samples. This innovative method utilizes the ability of methyl CpG binding domain (MBD) proteins to bind methylated DNA with high affinity.

Measure DNA methyltransferase activity or screen for enzyme inhibition

The DNMT Activity / Inhibition Assay provides all the reagents needed to study DNA methyltransferase activity from recombinant DNMT enzymes or nuclear extract samples (Figure 1). The sensitive ELISA-based method utilizes the high-affinity binding of a methyl CpG binding domain (MBD) to methylated DNA. In the DNMT assay method, a universal CpG-enriched DNA substrate has been immobilized on a 96-stripwell plate. Purified DNMTs or DNMT activities from nuclear extracts are added, which catalyze the transfer of methyl group to the DNA substrate. The resulting methylated DNA is recognized and bound by the recombinant MBD protein in a manner proportional to the enzyme's activity. The assay also includes a sample of purified DNMT1 enzyme as a positive control.

DNMT Assay advantages

- **Non-radioactive** – colorimetric assay is easily quantified on a microplate reader
- **Sensitive** – unique methyl CpG binding domain approach enhances sensitivity
- **Fast** – assay can be completed in less than 3 hours
- **Less effort required** – compatible with multi-channel pipettors to streamline wash steps
- **Flexible** – stripwell plate enables screening in low or high throughput

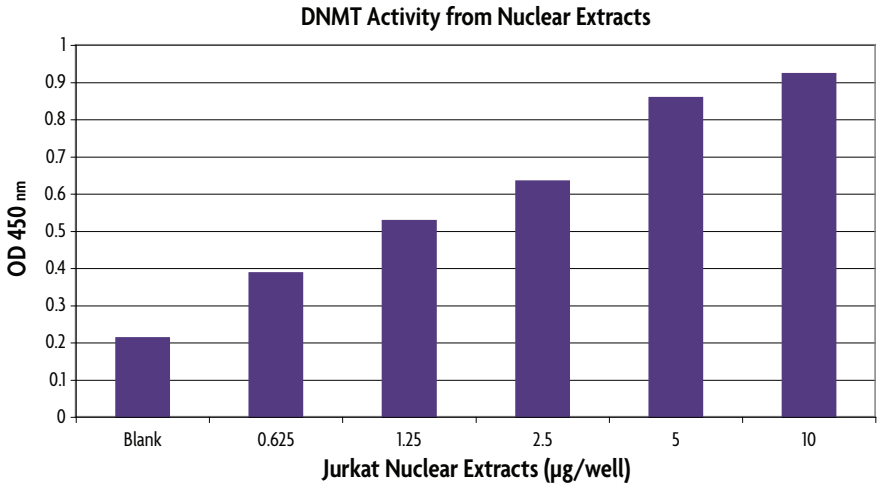

Figure 1: DNMT activity assayed from Jurkat nuclear extracts.
The DNMT Activity / Inhibition Assay was used to screen Jurkat nuclear extracts prepared using Active Motif's Nuclear Extract Kit (Catalog No. 40010). The assay can detect DNMT activity from as little as 0.625 µg of extract with a 1.5 hour incubation and a 5 minute developing time. Data shown are the results from wells assayed in duplicate.

Why study DNMTs?

In mammals and other vertebrates, DNA methylation occurs at the C5 position of cytosine (5-mC), mostly within CpG dinucleotides. Modification at these sequences by DNA methyltransferase enzymes (DNMTs) can have profound effects on transcription. DNA methylation can cause gene silencing by recruitment of repressors, such as methyl-CpG-binding domain (MBD) proteins, that recruit protein complexes to establish silent chromatin. The study of DNMTs and DNA methylation is relevant to many aspects of biology and disease.

Active Motif offers a wide variety of products to aid in your research of DNA methylation and DNA methyltransferase enzymes. These include recombinant enzymes, antibodies, kits to enrich for both methylated and unmethylated DNA and DNA standards for use as controls. For more information, please visit www.activemotif.com/dnamt.

Product	Format	Catalog No.
DNMT Activity / Inhibition Assay	1 x 96 rxns	55006